# FACSIMILE MESSAGE

**OUR FAX#** 732-445-0072
**OUR PHONE#** 732-445-4256

**TO:** Eric P. Mirabel
BioArray Solutions
35 Technology Drive
Suite 100
Warren, NJ 07059

Confidential!

**FAX:** 908-226-0800

**FROM:** Joachim Messing, Director
Waksman Institute
Rutgers, The State University of New Jersey
190 Frelinghuysen Road
Piscataway, NJ 08854-8020

**DATE:** February 17th, 2009 **PAGES TRANSMITTED:** 14

**MESSAGE:**

Dear Eric,

Please, find enclosed my declaration regarding Office Action of November 14th, 2008, related to U.S. Patent Application Serial No. 10/645,426. Please, confirm receipt via email to: messing@waksman.rutgers.edu.

Thank you,

Joachim Messing

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

| | | |
|---|---|---|
| In re Application of: Michael Seul | § | Group Art Unit: 1641 |
| | § | |
| | § | Examiner: Pensee T. Do |
| Serial No.: 10/645,426 | § | |
| | § | |
| Filed: **August 21, 2003** | § | |
| For: **LIGHT-CONTROLLED ELECTROKINETIC ASSEMBLY OF PARTICLES NEAR SURFACES** | § | |

February 15th, 2009

## DECLARATION OF DR. JOACHIM MESSING

## A. QUALIFICATIONS

1. I, Joachim Messing, have been asked to prepare a declaration on issues raised by the recent Office Action of November 14th, 2008, related to U.S. Patent Application Serial No. 10/645,426 (hereinafter "the '426 application").

2. My qualifications for providing this declaration are as follows. I received an M.S. degree in Pharmacy from the Free University of Berlin, graduate training at the Max-Planck Institute of Biochemistry, and a doctorate degree from the Ludwig Maximilian University of Munich. I held research positions at the Max-Planck Institute at Martinsried, and at the University of California, at San Francisco and Davis. Subsequently, I was a Professor of Biochemistry at the University of Minnesota and, in 1985, moved to Rutgers, The State

University of New Jersey, as a University Professor and Director of the Waksman Institute of Microbiology. Since then, I have founded two new departments at Rutgers, the Department of Molecular Biology and Biochemistry and the Department of Genetics, and served as their initial chair, in addition to my responsibilities at the Institute. My particular research interests are in the use of my early gene cloning techniques to study the molecular and genetic mechanisms of quantitative traits in plants for improving the nutritional quality of corn but I am knowledgeable and skilled in the art of methods in molecular biology. I am particularly well known for initiating and developing the use of single-stranded DNA phage M13, the pUC plasmids, universal primers and polycloning sites for gene cloning and DNA sequencing. These widely used techniques and strains have had a far-reaching impact on biotechnological research worldwide, which became clear in October 1991, when I was named the most frequently cited scientist in the decade (1981-1990). In addition, see my curriculum vitae in short form at Wikipedia and in detail as Exhibit 1.

3. After Dr. Seul founded BioArray Solutions (hereafter BAS) more than 10 years ago, he sought advice in the area of molecular biology and asked me to serve as the chair of his scientific advisory committee. I have subsequently visited his research group on a regular basis. I am therefore quite familiar with the implementation of his ideas in the use of BAS' bead array technology in bioassays, the experimentation and development performed over the years by BAS' employees, and the real life data from patients that have benefited from the application of this technology.

## B. COMPENSATION AND PREVIOUS CONSULTATION

4. BioArray Solutions is compensating me for my time spent on this matter at my usual consulting rate of $350.00 per hour. My compensation is not contingent on any outcome in the case.

5. In addition to this case, I am a consulting expert for Monsanto Co. on various intellectual property matters and I have testified as an expert for Monsanto Co. at trial, by deposition, and/or by declaration in these cases: (1) *Syngenta Seeds, Inc. v. Monsanto Co., DEKALB Genetics Corp., Pioneer Hi-Bred International, Inc., Dow Agrosciences, LLC, Mycogen Plant Science, Inc. and Agrigenetics, Inc., collectively d.b.a. Mycogen seeds*, Case No. 02-1331-SLR; (2) *Mycogen Plant Science, Inc. and Agrigenetics Inc. v. Monsanto Co.*, C.A No. 950653B (LSP), 96-505 RRM, 1:04-CV-0573 DFH-WTL; (3) *Monsanto Co. v. Aventis*, 4:000CV01915; (4) *Plant Genetic Systems, N.V. v. DEKALB Genetics Corporation*, 3:96CV2015; (5) *Johnston v. Beachy* (interference), Patent Interference No. 104,286; (6) *Pioneer Hi-Bred International v. Monsanto Company*, No. 4:97CV1609ERW; (7) re-examination of USP 5,352,605. I have also provided testimony as an expert in *Michigan State v. Peoples* (interference), but not for Monsanto Co.

## C. ONE OF ORDINARY SKILL IN THE ART

6. In my opinion, a person of ordinary skill in this art at the time of filing would be somebody with a Ph.D. in molecular biology or genetics with a few years of practical experience. This person likely would be working in a team of scientists and technicians with a range of areas of specialization such as genetics, biochemistry, cell and molecular biology, software developers, and electrical engineers, who are familiar with semiconductors.

## D. SUMMARY OF OPINION

7. I have reviewed the Office Action of the '426 application "Light-Controlled Electrokinetic Assembly of Particles near Surfaces of Dr. Michael Seul" by examiner Pensee Do on the written description argument according to 35 USC 112 that no sufficient information has been provided to a person skilled in the art to the amended language of claim 76 "*the particles are affixed to the substrate in a loosely packed, ordered array.*" The examiner in particular cited the first paragraph of 35 USC 112: "*The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.*" After careful consideration of the specification of the '426 application, I would like to take exception to the examiner's arguments and conclude that the specification fully support the amended claim. All citations below from the '426 application are highlighted in italics and referenced by page and line numbers.

## E. PACKING OF PARTICLES

8. The examiner quotes from the specification that "as a function of increasing applied voltage, beads first form planar aggregates in which particles are mobile and loosely packed, then assume a tighter packing, and finally exhibit a spatial arrangement in a form of a crystalline, or ordered array resembling a raft of bubbles" and concludes "the beads can either be loosely packed, or in an ordered array but never in both stages." Furthermore, the examiner rejects the interpretation of the specification that the beads undergo three transformations: "1)

mobile and loosely packed; 2) assume tighter packing; 3) spatial arrangement in an ordered array as the applied voltage increases" as new matter.

9. The definition of particles and their arrangements has been described in detail under "*Ions, Electric Fields and Fluid Flow: Field-induced Formation of Planar Bead Arrays*" on page 2 line 15 and visualized in several illustrations, which are described in detail under "*Detailed Description of the Preferred Embodiments*" on page 11, line 4.

10. To better understand the forces that apply to the motion and arrangement of particles, one has to read the reference cited on page 3, line 6, where it says: " *it has been only recently demonstrated that the underlying attractive interaction between beads is mediated by electrokinetic flow (Yeh, Seul and Shraiman, "Assembly of Ordered Colloidal Aggregates by Electric Field Induced Fluid Flow", Nature 386, 57-59 (1997).*" In particular, Figure 3 of this article is a good illustration of the forces that apply to particles as described in the invention. These forces, reflecting fundamental aspects of the competing forces underlying the array assembly process, guarantee that the constituent particles within a bead array, under the conditions specified for the realization of a planar crystalline arrangement, undergo different stages of mobility, packing, and pattern of arrangements. To deny that aspect of the invention would be to deny well established principles of physics and physical chemistry.

11. Please note, with respect to the Nature paper, that Dr. Seul's, and BioArray Solutions' address was that of my Institute where the early work of developing commercial applications of the technology was undertaken; as suggested by that fact, I had frequent contact with Dr Seul, and thus became quite familiar with the essential aspects of the random encoded bead array platform. At that time, Dr. Seul also received a government grant to develop applications of that format to molecular diagnostic tests: since then, and continuing to this day,

he and others at BioArray Solutions have worked diligently to successfully reduce his invention to practice. In due course, therefore, it has been proven that particles indeed can be arranged in different stages as described in the Specification.

12. After founding BioArray Solutions, Dr. Seul was able to show that "*the flow-mediated attractive interaction between beads extends to distances far exceeding the characteristic bead dimension*" (see page 3, line 24). Because the bead size could be measured and the arrays of beads visualized, the distances between adjacent beads were defined in these arrays, as I will show in more detail below. By controlling the strength of the electric field between the electrodes, distances between adjacent particles can be adjusted by tuning the relevant known parameters.

13. Continuing on page 3, line 26, the Specification says: "*Planar aggregates are formed in response to an externally applied electric field and disassemble when the field is removed. The strength of the applied field determines the strength of the attractive interaction that underlies the array assembly process and thereby selects the specific arrangement adopted by the beads within the array. That is, as a function of increasing applied voltage, beads first form planar aggregates in which particles are mobile and loosely packed, then assume a tighter packing, and finally exhibit a spatial arrangement in the form of a crystalline, or ordered array resembling a raft of bubbles.*" This language in the specification accurately addresses the three transformations of particle arrangements the examiner is referring to in the Office Action.

14. In addition to the electric field, the properties of the substrates play a critical role in array formation. For instance, as described in the specification, oxidation of silicon can be used to modify the substrate impedance (or more precisely the impedance of the interface between the oxide and the silicon). On page 4, line 20, the specification says: "*This modulation*

*in thickness, with typical variations of approximately 10 Angstroms, translates into spatial modulations in the impedance of the Si/SiOx interface while leaving a flat and chemically homogeneous top surface exposed to the electrolyte solution.*" This layer of oxidation - acting as an insulator - exemplifies the design of Electrolyte-Insulator-Semiconductor interface structures that have a well-defined effect on the electric field induced electrokinetic and photo-electrokinetic forces mediating array assembly. A critical aspect of the EIS structure is its effect on the effective spatial distribution of the interfacial electric field, which in turn determines the movement of beads under the influence of AC electric fields and light, providing a method of generating local field gradients and corresponding forces in specific areas of a silicon substrate (e.g. compare Fig. 4a and 4b). Light absorbed by these substrates acts to modulate the local interfacial impedance.

15. The beauty of this concept is that it introduces the use and the low cost of wafers to solid-support biochemistry, an important field in medical diagnostics and molecular biology in general. Furthermore, because beads can be chemically labeled, wafers can be used to miniaturize, highly parallelize, and monitor many different biochemical reactions. As the Specifications says in Example V on page 26, line 19: "*The present method relies on the functional elements of the invention to assemble a planar array of a multi-component mixture of beads which carry chemical labels in the form of tag molecules and may be so identified subsequent to performing the assay.*"

16. The operational qualities of these bead arrays is then summarized under the "*Summary of the Invention*" on page 8, line 32: "*The combination of three functional elements endows the present invention with a set of operational capabilities to manipulate beads and bead arrays in a planar geometry to allow the implementation of biochemical analytical techniques.*"

## F. DOCUMENTATION

17. The "Detailed Description of the Preferred Embodiments" provides a person skilled in the art the information as to how, starting from a standard wafer, to build the appropriate semiconductor substrate. It also describes how beads can be formed and modified, placed on the semiconductor, and arrayed. It provides the exact dimensions of the semiconductor and the colloidal beads. Actually, the system has a strikingly simple configuration and components because it can use off-the-shelve materials. All that is needed is to register and record the movement and aggregation of beads is a microscope. By attaching a simple CCD device (camera) to the microscope, pictures of arrays can be recorded as shown in the Nature publication and the drawings of the specification.

18. A critical step in controlling the distance of adjacent beads is the number of beads placed in the electrochemical cell. On page 11, line 22, the Specification states: "*The cell is first assembled and then filled, relying on capillary action, with a suspension of colloidal beads, 1 or 2 microns in diameter, at a typical concentration of 0.1 % solids in 0.1 mM azide solution, corresponding to approximately 2 x $10^8$ particles per milliliter. The number is chosen so as to yield between 1/2 and 1 full monolayer of particles on the electrode surface.*" As one can see, monolayer coverage is achieved empirically by being able to see the beads and their distances directly under the microscope (see page 12, line 30).

19. As one can see in Figure 2 of the Specification, the photograph of 2a shows no electric field and no bead capture, while 2b provides an example in what is called "*region 22*," where beads are captured due to an electric field. As the Specification states on page 12, line 22: "*The internal state of order of the captured aggregate of beads is determined by the strength of*

*the applied voltage, higher values favoring increasingly denser packing of beads and the eventual formation of ordered arrays displaying a hexagonally crystalline configuration in the form of a bubble raft.*" The regions 24 and 26 in Figure 2d illustrate the influence of the thickness of the SiOx layer on impedance as discussed already above. Clearly, the three transformations of particle arrangements are not new matter, but documented with examples.

20. A striking property of this system is the use of light, as the title of the invention says, to create differently packed particles in different channels on the same slide as shown in Figure 3, which produces multiple rows of arrays on a single flat surface. The reason for this is that a light beam can be targeted and pulsed to specific areas of a planar substrate surface. Therefore, the method of the invention can induce bead transport, including array translocation, and can generate temporary aggregates of beads. As the Specification says on page 14, line 16: "*The present invention provides for mechanisms of light-mediated active linear transport of planar aggregates of beads under interactive control.*" This level of manipulation of arraying beads clearly illustrates that a person skilled in the art can control movement and distance of adjacent beads by setting relevant parameters. Because that person would be able to instantly record patterns of assembly and planar arrays of beads by microscopy, various conditions could also be achieved empirically without undue experimentation. Furthermore, beads could be arrayed in a flexible manner dependent on the application the person skilled in the art would like to exercise.

21. The precision, with which arrays can be formed and controlled, is also illustrated in Figure 4. Beads captured in region O in Figure 4a are split in subarrays F1, F2, and Fn as shown in Figure 4b, illustrating the formation of multiple rows of arrays. In each subarray, adjacent beads have about the same distance from each other as can be seen in the photograph.

As the specification says on page 16, line 15: "*This fragmentation of an array into smaller clusters reflects the effect of a field-induced particle polarization. The splitting is useful to distribute particles in an array over a wider area of substrate for presentation to possible analytes in solution, and for subsequent scanning of the individual clusters with analytical instruments to make individual readings.*" Note that here anionic, carboxylated polystyrene beads of 2-micron diameter - roughly the length of an *E. coli* bacterium, which also can be seen under the microscope - are used as described on page 18, line 22, in more detail.

## F. DEMONSTRATIONS

22. Additional details regarding experimental conditions can be found in the Specification under "General Experimental Conditions," which applies to all the examples. It instructs the person skilled in the art to keep the concentration of beads to a level not to produce more than a complete monolayer of particles (page 18, line 19). Once a monolayer is formed, the Specification notes on page 20, line 12: "*The present invention provides a method of forming planar arrays with precise control over the mechanical, optical, and chemical properties of the newly created layer.*"

23. Furthermore, it notes that these properties provide the person skilled in the art advantages over prior art: "*The process of the present invention enables the formation of ordered planar arrays from the liquid phase (in which particles are originally suspended) in designated positions, and in accordance with a given overall outline. This eliminates the above-stated disadvantages of the prior art, i.e., dry state, irregular or no topography, random placement within an aggregate, immobilization of particles and uncontrolled, random placement of ordered*

*patches on the substrate*." Critical advances are the speed and precision with which these arrays can be formed using these properties.

24. Although such a layer represents a (compositionally) random distribution of beads, in the event that beads of distinguishably different properties (for example: different chemical functionalities) are used, each bead occupies a position, within the planar array, that is well defined with respect to its neighbors, and can be controlled with respect to location and distance to those neighbors. As the Specification says under Example I: "*In this way, the location of the individual beads is random, but the relative proportion of each type of bead within the array is controllable.*"

25. In Example II, the Specification emphasizes the nature of planar arrays to reflect "crystalline" order: "*The present invention provides for a rapid and well controlled process of forming planar arrays in a state of crystalline order, which will function as surface-mounted optical diffraction elements.*" Crystalline order implies the distance between beads within the array to be identical, as claimed. Furthermore, the Specification notes the advantages of the present invention in achieving such crystalline: "*In contrast to the slow and cumbersome prior art method of fabricating such arrays by way of forming equilibrium crystals in aqueous solutions of low salt content, the present invention provides a novel approach to rapidly and reliably fabricate particle arrays at a solid-liquid interface.*"

26. Example V, which describes applications of arrays to diagnostic tests, also highlights properties of these arrays and their unique features. The Specification on page 28, line 5 says: "*In contrast to all prior art methods, the present invention provides a novel method to create heterogeneous panels by in-situ, reversible formation of a planar array of "encoded" beads in solution adjacent to an electrode. The array may be random with respect to chemical*

*identity but is ordered with respect to spatial position.*" Without these properties of arranging particles in an ordered planar array, one could not have achieved the present-day application of this invention to the extensive determination of antigens on red blood cells, that is, the determination of extended red cell ("blood") types of thousands of patients.

27. Demonstration of these properties also is presented in Figure 7 which not only shows that in each subfield, 72 and 74, adjacent particles are spatially arranged in such a way that it can vary in each subfield. As the Specification says on page 28, line 16: "*Third, to accommodate scanning probe analysis of individual beads, interparticle distances within the array may be adjusted by field-induced polarization or by the addition of inert spacer particles that differ in size from the encoded beads. Figure 7 shows the use of small spacer beads 72 for separating encoded beads 74. As shown, the spacing of beads 74 is greater than the spacing of comparable beads in Figure 4b.*"

28. The formation of these types of arrays of beads is critical for the application of the invention in diagnostic tests because of the encoding step. The Specification notes on page 28, line 28: "*This novel approach of in-situ assembly of panels relies on beads that carry a unique chemical label, or code, to permit their identification subsequent to the completion of a binding assay. This invention facilitates on-line tagging of beads by way of a photochemical bead coloring method.*"

29. Multiple rows also can be created by the "Layout-Preserving Transfer" method. As the Specification says on page 44, line 2: "*As with heterogeneous panels in general, the arrangement of beads within the array is either random (with respect to chemical identity), and the identity of beads scoring high in the binding assay must be determined subsequently, or it is spatially encoded by invoking the "Layout-Preserving Transfer" method of sample loading*

*described herein.*" Therefore, different stages of packing particles can be achieved on the same surface.

30. Example VII further addresses this: as described above, light-induced movement can generate spatially ordered configurations of (compositionally) randomly distributed beads. As the Specification says on 37, line 22: "*The invention may be used to induce the formation of particle chains in the direction normal to the plane of the electrode. The chains represent conduits for current transport between the electrodes and their formation may reflect a field-induced polarization. Chains are much less mobile in transverse flow than are individual particles so that this effect may be used to separate particles according to the surface properties that contribute to the net polarization. The effect of reversible chain formation has been demonstrated under the experimental conditions stated herein. For example, the reversible formation of chains occurs, for carboxylated polystyrene beads of 1 micron diameter, at a voltage of 15 V (PP) at frequencies in excess of 1 MHz.*" By creating these chains, multiple rows of arrays can be formed on the same planar surface.

I make the foregoing statements having understood that willful false statements and the like are punishable by fine or imprisonment, or both, and may jeopardize the validity of the application or any patent issuing thereon.

February 17th, 2009
Date

Joachim Messing